

We claim:

Claim 1. A method for the treatment of a particular volume of tissue, said volume of tissue containing an endogenous pigment, the method comprising the steps of:  
treating the particular volume of tissue with light to promote a simultaneous two-photon photoactivation of said pigment in the particular volume of tissue, wherein the pigment becomes photochemically activated in the particular volume of tissue.

Claim 2. The method of Claim 1 wherein the light to promote said simultaneous two-photon photoactivation is a laser light produced by a laser.

Claim 3. The method of Claim 2 wherein the laser light comprises a train of one or more ultrashort pulses.

Claim 4. The method of Claim 2 including operating the laser to produce light at a wavelength between approximately 450 nm to 1400 nm.

Claim 5. The method of Claim 1 wherein the light to promote said simultaneous two-photon photoactivation is a focused beam of light.

Claim 6. The method of Claim 5 wherein the focused beam of light is focused laser light.

Claim 7. The method of Claim 6 wherein said particular volume of tissue is located substantially at the tissue surface.

**Claim 8.** The method of Claim 6 wherein said particular volume of tissue is located substantially below the tissue surface.

5 **Claim 9.** The method of Claim 1 wherein said step of treating the particular volume of tissue includes positioning a focus of a beam of light over a range of positions so that a focal plane of the light beam occurs at a site located between a surface of the tissue and a point substantially beyond the tissue surface, whereby said step of treating the particular volume of tissue may extend to penetrate deep within the tissue.

10 **Claim 10.** The method of Claim 9 further including varying, while the beam of light is extant, the radial position of the focal plane within the tissue, thereby to photoactivate the endogenous pigment at a multiplicity of positions between the tissue surface and a position located substantially beyond the tissue surface.

15 **Claim 11.** The method of Claim 1 wherein said endogenous pigment becomes photoactivated in said particular volume at a controllable position substantially beyond a tissue surface.

20 **Claim 12.** The method of Claim 1 further comprising the step of controlling the photoactivation by varying the location, irradiance and duration of said light.

**Claim 13.** The method of Claim 1 wherein the light to promote said simultaneous two-photon excitation of the endogenous pigment is a non-focused beam of light.

Claim 14. The method of Claim 13 wherein said particular volume of tissue is located substantially at the tissue surface.

5 Claim 15. The method of Claim 13 wherein said particular volume of tissue is located substantially below the tissue surface.

Claim 16. The method of Claim 1 wherein said endogenous pigment is selected from the group comprising melanin, melanin precursors, carotenes, porphyrins, and various tattoo dyes.

10 Claim 17. The method of Claim 16 wherein said melanin precursors are selected from the group comprising 5-S-cysteinyldopa (5-SCD) and 5,6-dihydroxyindole (DHI), dopa, dopa semiquinone, leucodopachrome, dopachrome, eumalanins, pheomelanins, sepia melanins, and 5,6-dihydroxyindole-2-carboxylic acid.

15 Claim 18. The method of Claim 16 wherein said porphyrins include hemoglobin.

20 Claim 19. A method for producing a photoactivated product in a particular volume of a material, the method comprising treating the particular volume of the material with light to promote a simultaneous two-photon excitation of an endogenous pigment contained in the particular volume of the material, wherein the pigment becomes a photoactivated product in the particular volume of the material.

Claim 20. The method of Claim 19 wherein the light to promote said simultaneous two-photon photoactivation is a laser light produced by a laser.

Claim 21. The method of Claim 20 wherein the laser light comprises a train of one or more ultrashort pulses.

Claim 22. The method of Claim 20 including operating the laser to produce light at a wavelength between approximately 450 nm to 1400 nm.

Claim 23. The method of Claim 19 wherein the light to promote said simultaneous two-photon photoactivation is a focused beam of light.

Claim 24. The method of Claim 23 wherein the focused beam of light is focused laser light.

Claim 25. The method of Claim 24 wherein said particular volume of material is tissue located substantially at the surface of said material.

Claim 26. The method of Claim 24 wherein said particular volume of material is tissue located substantially below the surface of said material.

Claim 27. The method of Claim 19 wherein said step of treating the particular volume of material includes positioning a focus of a beam of light over a range of positions so that a focal plane of the light beam occurs at a site located between a surface of the

material and a point substantially beyond the material surface, whereby said step of treating the particular volume of material may extend to penetrate deep within the material.

5      Claim 28. The method of Claim 27 further including varying, while the beam of light is extant, the radial position of the focal plane within the material, thereby to photoactivate the endogenous pigment at a multiplicity of positions between the material surface and a position located substantially beyond the material surface.

10      Claim 29. The method of Claim 19 wherein said endogenous pigment becomes photoactivated in said particular volume at a controllable position substantially beyond a material surface.

15      Claim 30. The method of Claim 19 further comprising the step of controlling the photoactivation by varying the location, irradiance and duration of said light.

20      Claim 31. The method of Claim 19 wherein the light to promote said simultaneous two-photon excitation of the endogenous pigment is a non-focused beam of light.

25      Claim 32. The method of Claim 31 wherein said particular volume of material is located substantially at the surface of said material.

30      Claim 33. The method of Claim 31 wherein said particular volume of material is tissue located substantially below the surface of said material.

Claim 34. The method of Claim 19 wherein said endogenous pigment is selected from the group comprising melanin, melanin precursors, carotenes, porphyrins, and various tattoo dyes.

5        Claim 35. The method of Claim 34 wherein said melanin precursors are selected from the group comprising 5-S-cysteinyldopa (5-SCD) and 5,6-dihydroxyindole (DHI), dopa, dopa semiquinone, leucodopachrome, dopachrome, eumalanins, pheomelanins, sepia melanins, and 5,6-dihydroxyindole-2-carboxylic acid.

10        Claim 36. The method of Claim 34 wherein said porphyrins include hemoglobin.

15        Claim 37. A method for treatment of tissue wherein the tissue includes an endogenous pigment, the method comprising the steps of:

20        directing light to specific regions of interest within the tissue, including regions substantially below a tissue surface, said light being selected to penetrate the tissue and to promote two-photon excitation substantially only at a focal zone;

      controlling the location of said focal zone over a range of depths within said tissue; and

      using two-photon excitation, photoactivating said pigment over said range of depths within said tissue, thereby producing a photoactivated product substantially only at the focal zone.

Claim 38. The method of Claim 37 wherein said directing step includes directing a laser light produced by a laser to said regions of interest.

Claim 39. The method of Claim 38 wherein the laser light comprises a train of one or more ultrashort pulses.

Claim 40. The method of Claim 38 including operating the laser to produce light at a wavelength between approximately 450 nm to 1400 nm.

Claim 41. The method of Claim 37 wherein the light to promote said two-photon photoactivation is a focused beam of light.

Claim 42. The method of Claim 41 wherein the focused beam of light is focused laser light.

Claim 43. The method of Claim 42 wherein said regions of interest are located substantially at the tissue surface.

Claim 44. The method of Claim 42 wherein said regions of interest are located substantially below the tissue surface.

Claim 45. The method of Claim 42 further comprising the step of scanning said regions of interest with said focused beam of light to promote two-photon excitation throughout said regions of interest.

Claim 46. The method of Claim 37 wherein said endogenous pigment becomes photoactivated in said focal zone at a controllable position substantially beyond a tissue surface.

5        Claim 47. The method of Claim 37 wherein said two-photon photoactivation is simultaneous two-photon activation.

Claim 48. The method of Claim 37 further comprising the step of controlling the photoactivation by varying the location, irradiance and duration of said light.

10        Claim 49. The method of Claim 37 wherein the light to promote said two-photon excitation of the photoactive agent is a non-focused beam of light.

15        Claim 50. The method of Claim 49 wherein said regions of interest are located substantially at the tissue surface.

Claim 51. The method of Claim 49 wherein said regions of interest are located substantially below the tissue surface.

20        Claim 52. The method of Claim 37 wherein said endogenous pigment is selected from the group comprising melanin, melanin precursors, carotenes, porphyrins, and various tattoo dyes.



Claim 53. The method of Claim 52 wherein said melanin precursors are selected from the group comprising 5-S-cysteinyldopa (5-SCD) and 5,6-dihydroxyindole (DHI), dopa, dopa semiquinone, leucodopachrome, dopachrome, eumalanins, pheomelanins, sepia melanins, and 5,6-dihydroxyindole-2-carboxylic acid.

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Claim 54. The method of Claim 52 wherein said porphyrins include hemoglobin.

Claim 55. A method for the treatment of a particular volume of tissue, said volume of tissue containing an endogenous pigment, the method comprising the steps of:

10 treating the particular volume of tissue with light to promote thermal overload of pigmented <sup>cells</sup> in the particular volume of tissue, wherein said thermal overload kills said pigmented cells.

15 Claim 56. The method of Claim 55 wherein the light to promote said thermal overload is a laser light produced by a laser.

Claim 57. The method of Claim 56 wherein the laser light comprises a train of one or more ultrashort pulses.

20 Claim 58. The method of Claim 56 including operating the laser to produce light at a wavelength between approximately 450 nm to 800 nm.

Claim 59. The method of Claim 58 wherein said wavelength of light is between approximately 600 nm and 800 nm.

Claim 60. The method of Claim 58 wherein said particular volume of tissue is located substantially at the tissue surface.

Claim 61. The method of Claim 58 wherein said particular volume of tissue is located approximately 2 mm or less below the tissue surface.

Claim 62. The method of Claim 58 wherein said laser light has a pulse duration of less than 10 ns.

Claim 63. The method of Claim 62 wherein said laser light has a pulse duration of less than 10 ps.

Claim 64. The method of Claim 56 including operating the laser to produce light at a wavelength between approximately 800 nm to 1400 nm.

Claim 65. The method of Claim 64 wherein said particular volume of tissue is located approximately 2 mm or greater below the tissue surface.

Claim 66. The method of Claim 64 wherein said laser light has a pulse duration of less than 10 ps.

Claim 67. The method of Claim 66 wherein said laser light has a pulse duration of less than 1 ps.

Claim 68. The method of Claim 55 wherein the light to promote said thermal overload is a focused beam of light.

Claim 69. The method of Claim 68 wherein the focused beam of light is focused  
5 laser light.

Claim 70. The method of Claim 68 wherein said step of treating the particular volume of tissue includes scanning said particular volume of tissue with said focused beam of light so as to promote thermal overload throughout said particular volume of tissue.

Claim 71. The method of Claim 55 wherein the light to promote said thermal overload is a non-focused beam of light.

Claim 72. The method of Claim 55 wherein said endogenous pigment is selected  
15 from the group comprising melanin, melanin precursors, carotenes, porphyrins, and various tattoo dyes.

Claim 73. The method of Claim 72 wherein said melanin precursors are selected from the group comprising 5-S-cysteinyl-dopa (5-SCD) and 5,6-dihydroxyindole (DHI), dopa,  
20 dopa semiquinone, leucodopachrome, dopachrome, eumelanins, pheomelanins, sepia melanins, and 5,6-dihydroxyindole-2-carboxylic acid.

Claim 74. The method of Claim 72 wherein said porphyrins include hemoglobin.

Claim 75. A method for treatment of a particular volume of tissue, said volume of tissue containing an endogenous pigment and an exogenous photodynamic agent, the method comprising the steps of:

treating the particular volume of tissue with light to promote a simultaneous two-photon photoactivation of said pigment and said agent in said particular volume of tissue, wherein the pigment becomes photochemically converted into a phototoxic product in the particular volume of tissue and said photodynamic agent becomes photoactivated in the particular volume of tissue.

Claim 76. The method of Claim 75 wherein said exogenous photodynamic agent is selected from the group comprising Rose Bengal, psoralen derivatives, indocyanine, Lutex,  $\text{Sn(ET)}_2$ , and various porphyrin derivatives, including porfimer sodium and benzoporphyrin derivative.

Claim 77. The method of Claim 75 wherein the particular volume of tissue is pretreated with said exogenous photodynamic agent such that the particular volume of tissue retains a portion of said agent at the time the particular volume of tissue is treated with light so as to promote simultaneous two-photon activation of said agent.

Claim 78. Apparatus for treating a particular volume of tissue containing an endogenous pigment, the apparatus comprising:

a source of light and light delivery apparatus for directing light at and into said particular volume of tissue, said light being selected in frequency and energy to promote

simultaneous two-photon excitation of said endogenous pigment so that said pigment becomes photochemically activated in said particular volume of tissue.

Claim 79. The apparatus of Claim 78 wherein said endogenous pigment is selected from the group comprising melanin, melanin precursors, carotenes, porphyrins, and various tattoo dyes.

Claim 80. The apparatus of Claim 79 wherein said melanin precursors are selected from the group comprising 5-S-cysteinyl-dopa (5-SCD) and 5,6-dihydroxyindole (DHI), dopa, dopa semiquinone, leucodopachrome, dopachrome, eumalanins, pheomelanins, sepia melanins, and 5,6-dihydroxyindole-2-carboxylic acid.

Claim 81. The apparatus of Claim 79 wherein said porphyrins include hemoglobin.

Claim 82. The apparatus of Claim 79 wherein said source of light is a laser light produced by a laser.

Claim 83. The apparatus of Claim 82 wherein said laser light comprises a train of one or more ultrashort pulses.

Claim 84. The apparatus of Claim 82 wherein said laser light has a wavelength between approximately 450 nm to 1400 nm.

Claim 85. The apparatus of Claim 78 wherein said particular volume of tissue is located substantially at the tissue surface.

Claim 86. The apparatus of Claim 78 wherein said particular volume of tissue is located substantially below the tissue surface.

Claim 87. The apparatus of Claim 78 wherein said light is non-focused light.

Claim 88. The apparatus of Claim 78 further comprising a focusing apparatus for focusing the light throughout a range of focal lengths extending from a surface of said tissue to a depth substantially beyond said surface, said source of light and focusing apparatus cooperating to promote simultaneous two-photon excitation of said pigment.

Claim 89. The apparatus of Claim 78 further comprising an exogenous photodynamic agent in said particular volume of tissue, said light being selected in frequency and energy to promote simultaneous two-photon activation of said agent so that said agent becomes photoactivated in the particular volume of tissue.

Claim 90. The apparatus of Claim 89 wherein said exogenous photodynamic agent is selected from the group comprising Rose.Bengal, psoralean, indocyanine, Lutex,  $\text{Sn(ET)}_2$  and various porphyrin derivatives, including porfimer sodium and benzoporphyrin derivative.

Claim 91. An apparatus for treating a particular volume of tissue containing an endogenous pigment, the apparatus comprising:

a source of light and light delivery apparatus for directing light at and into said particular volume of tissue, said light being selected to promote thermal overload of pigmented cells in the particular volume of tissue, wherein said thermal overload kills said pigmented cells.

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Claim 92. The apparatus of Claim 91 wherein said source of light is a laser light produced by a laser.

Claim 93. The apparatus of Claim 92 wherein said laser light comprises a train of one or more ultrashort pulses.

Claim 94. The apparatus of Claim 92 wherein said laser light has a wavelength between approximately 450 nm to 1400 nm.

Claim 95. The apparatus of Claim 91 wherein said particular volume of tissue is located substantially at the tissue surface.

Claim 96. The apparatus of Claim 91 wherein said particular volume of tissue is located substantially below the tissue surface.

Claim 97. The method of Claim 1 further comprising the step of heating said volume of tissue using said light so to produce a hyperthermic effect and controlling the hyperthermic effect by varying the location, irradiance and duration of said light so as to augment the effectiveness of said photoactivation.

Claim 98. The method of Claim 19 further comprising the step of heating said volume of material using said light so to produce a hyperthermic effect and controlling the hyperthermic effect by varying the location, irradiance and duration of said light so as to augment the effectiveness of said photoactivation.

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Claim 99. The method of Claim 1 wherein said photochemical activation of said pigment includes conversion of said pigment into a phototoxic product.

Claim 100. The method of Claim 1 wherein said photochemical activation of said pigment includes photobleaching of the pigment in said tissue.

Claim 101. The method of Claim 100 wherein said tissue is selected from the group comprising moles, freckles, hair follicles and tattoos.

Claim 102. The method of Claim 19 wherein said photoactivated product is a phototoxic product.

Claim 103. The method of Claim 19 wherein said photoactivation of said pigment includes photobleaching of the pigment in said material.

Claim 104. The method of Claim 103 wherein said material is selected from the group comprising moles, freckles, hair follicles and tattoos.



Claim 105. The method of Claim 37 wherein said photoactive product is a phototoxic product.

Claim 106. The method of Claim 37 wherein said photoactivating of said pigment includes photobleaching of said pigment in said tissue.

Claim 107. The method of Claim 106 wherein said tissue is selected from the group comprising moles, freckles, hair follicles and tattoos.

Claim 108. the apparatus of Claim 78 wherein said photochemical activation of said pigment includes conversion of said pigment into a phototoxic product.

Claim 109. The apparatus of Claim 78 wherein said photochemical activation of said pigment includes photobleaching of the pigment in said tissue.

Claim 110. The apparatus of Claim 109 wherein said tissue is selected from the group comprising moles, freckles, hair follicles, and tattoos.